Mycobacterium tuberculosis Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 2

Catalog No. NR-19638

This reagent is the tangible property of the U.S. Government.

For research use only. Not for human use.

Contributor:

Pathogen Functional Genomics Resource Center at the J. Craig Venter Institute

Manufacturer:

BEI Resources

Product Description:

Production in the 96-well format has increased risk of cross-contamination between adjacent wells. Individual clones should be purified (e.g. single colony isolation and purification using good microbiological practices) and sequence-verified prior to use. BEI Resources does not confirm or validate individual mutants provided by the contributor.

The *Mycobacterium tuberculosis (M. tuberculosis)* Gateway® clone set consists of 42 plates which contain 3724 sequence validated clones (3294 *M. tuberculosis,* strain H37Rv clones supplemented with 430 unique open reading frames (ORF) from *M. tuberculosis,* strain CDC1551) cloned in *Escherichia coli* (*E. coli*) DH10B-T1 cells. Each ORF was recombined in vector pDONR™221 with an ATG start codon and no stop codon. The sequence was validated by full length sequencing of each entry clone with greater than 1X coverage and a mutation rate of less than 0.2%. Detailed information about each clone is shown in Table 1.

Information related to the use of Gateway® Clones can be obtained from Invitrogen™. Recombination was facilitated through an *att*B substrate (*att*B-PCR product or a linearized *att*B expression clone) with an *att*P substrate (pDONR™221) to create an *att*L-containing entry clone. The entry clone contains recombinational cloning sites, *att*L1 and *att*L2 to facilitate gene transfer into a destination vector, M13 forward and reverse priming sites for sequencing and a kanamycin resistance gene for selection. Please refer to the Invitrogen™ Gateway® Technology Manual for additional details.

Plate orientation and viability were confirmed for NR-19638.

Material Provided:

Each inoculated well of the 96-well plate contains approximately 60 µL of *E. coli* culture (strain DH10B-T1) in Luria Bertani (LB) broth containing 50 µg/mL kanamycin supplemented with 15% glycerol.

Packaging/Storage:

NR-19638 was packaged aseptically in a 96-well plate. The product is provided frozen and should be stored at -80°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

Growth Conditions:

Media:
LB broth or agar containing 50 µg/mL kanamycin.
Incubation:
Temperature: *E. coli*, strain DH10B-T1 clones should be grown at 37°C.
Atmosphere: Aerobic
Propagation:

1. Scrape top of frozen well with a pipette tip and streak onto agar plate.
2. Incubate the plates at 37°C for 1 day.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Mycobacterium tuberculosis* Gateway® Clone Set, Recombinant in *Escherichia coli*, Plate 2, NR-19638."

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at www.beiresources.org.

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Use Restrictions:

This material is distributed for internal research, non-commercial purposes only. This material, its product or its

References:

1. Cole, S. T., et al. "Deciphering the Biology of *Mycobacterium tuberculosis* from the Complete Genome Sequence." Nature 393 (1998): 537-544. PubMed: 9634230.
2. Camus, J. C., et al. "Re-Annotation of the Genome Sequence of *Mycobacterium tuberculosis* H37Rv." Microbiology 148 (2002): 2967-2973. PubMed 12368430.

Support Provided by NIAID

Table 1: *Mycobacterium tuberculosis*, Gateway® Clones, Plate 2 (ZMTDB)[1]

Clone	Well Position	ORF Length	Locus ID	Description	Accession Number	Average Depth of Coverage
71363	A01	349	Rv1102c	hypothetical protein Rv1102c	NP_215618.1	2
71397	A02	349	Rv1772	hypothetical protein Rv1772	NP_216288.1	2
71322	A03	349	Rv1953	hypothetical protein Rv1953	NP_216469.1	1.905444126
71489	A04	349	Rv2809	hypothetical protein Rv2809	NP_217325.1	2
71466	A05	349	Rv3440c	hypothetical protein Rv3440c	NP_217957.1	2
71351	A06	349	Rv3615c	hypothetical protein Rv3615c	NP_218132.1	2
71431	A07	349	Rv3865	hypothetical protein Rv3865	NP_218382.1	2.191977077
71590	A08	349	Rv3905c	hypothetical protein Rv3905c	NP_218422.1	2
71378	A09	367	Rv0030	hypothetical protein Rv0030	NP_214544.1	2
71485	A10	370	Rv3129	hypothetical protein Rv3129	YP_177933.1	2
71285	A11	370	Rv3216	acetyltransferase	NP_217732.1	2
71297	A12	385	Rv0801	hypothetical protein Rv0801	NP_215316.1	2
71414	B01	385	Rv1466	hypothetical protein Rv1466	NP_215982.1	1.994805195
71294	B02	394	Rv0253	nitrite reductase	NP_214767.1	-
71263	B03	397	Rv1767	hypothetical protein Rv1767	NP_216283.1	2.549118388
71518	B04	400	Rv1089	PE family protein	YP_177785.1	-
71602	B05	400	Rv2658c	prophage protein	NP_217174.1	2
71618	B06	400	Rv3922c	hypothetical protein Rv3922c	NP_218439.1	-
71502	B07	403	Rv1311	F0F1 ATP synthase subunit epsilon	NP_215827.1	1.990074442
71575	B08	406	Rv0662c	hypothetical protein Rv0662c	NP_215176.1	1.884236453
71506	B09	406	Rv0714	50S ribosomal protein L14	NP_215228.1	2
71353	B10	406	Rv2087	hypothetical protein Rv2087	NP_216603.2	2
71471	B11	412	Rv2206	transmembrane protein	NP_216722.2	1.601941748
71585	B12	417	Rv0857	hypothetical protein Rv0857	NP_215372.2	1.932853717
71254	C01	421	Rv1761c	hypothetical protein Rv1761c	NP_216277.1	2.589073634
71317	C02	422	Rv2863	hypothetical protein Rv2863	NP_217379.1	2
71342	C03	424	Rv2898c	hypothetical protein Rv2898c	NP_217414.1	2
71543	C04	427	Rv0367c	hypothetical protein Rv0367c	NP_214881.1	1.606557377
71433	C05	427	Rv1052	hypothetical protein Rv1052	NP_215568.1	1.927400468
71439	C06	427	Rv2570	hypothetical protein Rv2570	NP_217086.1	1.995316159
71637	C07	493	Rv0985c	large-conductance mechanosensitive channel	NP_215500.1	2
71682	C08	502	Rv2576c	hypothetical protein Rv2576c	NP_217092.1	2
72342	C09	514	Rv1276c	hypothetical protein Rv1276c	NP_215792.1	2.342412451
72197	C10	514	Rv2239c	hypothetical protein Rv2239c	NP_216755.1	2
72186	C11	514	Rv2730	hypothetical protein Rv2730	NP_217246.1	2
72351	C12	514	Rv3437	transmembrane protein	NP_217954.1	2
72178	D01	517	Rv0374c	carbon monoxide dehydrogenase small subunit	NP_214888.1	1.998065764
72278	D02	517	Rv1926c	immunogenic protein MPT63 (antigen MPT63/MPB63) (16 kDa immunoprotective extracellular protein)	NP_216442.1	1.620889749

Clone	Well Position	ORF Length	Locus ID	Description	Accession Number	Average Depth of Coverage
72017	D03	517	Rv3581c	2-C-methyl-D-erythritol 2,4-cyclodiphosphate synthase	NP_218098.1	2
72026	D04	520	Rv0307c	hypothetical protein Rv0307c	NP_214821.1	1.907692308
72101	D05	520	Rv3165c	hypothetical protein Rv3165c	NP_217681.1	2
72042	D06	520	Rv3172c	hypothetical protein Rv3172c	NP_217688.1	2
72117	D07	520	Rv3237c	hypothetical protein Rv3237c	NP_217754.1	2
72135	D08	520	Rv3492c	Mce associated protein	NP_218009.1	2.380769231
72242	D09	520	Rv3831	hypothetical protein Rv3831	NP_218348.1	2.130769231
72023	D10	523	Rv1287	hypothetical protein Rv1287	NP_215803.1	2.361376673
72238	D11	523	Rv2633c	hypothetical protein Rv2633c	NP_217149.1	2
72327	D12	523	Rv2965c	phosphopantetheine adenylyltransferase	NP_217481.1	2.223709369
72158	E01	526	Rv0245	oxidoreductase	NP_214759.1	1.998098859
72115	E02	526	Rv2035	hypothetical protein Rv2035	NP_216551.1	2.933460076
72306	E03	529	Rv2327	hypothetical protein Rv2327	NP_216843.1	2.737240076
72378	E04	532	Rv0431	putative tuberculin related peptide	NP_214945.1	1.859022556
72191	E05	535	Rv2719c	hypothetical protein Rv2719c	NP_217235.1	1.439252336
72357	E06	538	Rv1506c	hypothetical protein Rv1506c	NP_216022.1	2
72053	E07	538	Rv2616	hypothetical protein Rv2616	NP_217132.1	2
72374	E08	541	Rv0201c	hypothetical protein Rv0201c	NP_214715.1	1.754158965
72273	E09	568	Rv3425	PPE family protein	YP_177971.1	2
72009	E10	576	Rv2987c	isopropylmalate isomerase small subunit	NP_217503.1	2
72171	E11	583	Rv0733	adenylate kinase	NP_215247.1	2.123499142
72361	E12	583	Rv1105	hypothetical protein Rv1105		2
72173	F01	589	Rv1503c	hypothetical protein Rv1503c	NP_216019.1	2
72249	F02	592	Rv1150	possible transposase fragment		-
72097	F03	595	Rv2499c	oxidase regulatory-like protein	NP_217015.1	1.996638655
72073	F04	598	Rv1476	hypothetical protein Rv1476	NP_215992.1	1.996655518
72067	F05	607	Rv1727	hypothetical protein Rv1727	NP_216243.1	-
72093	F06	610	Rv1388	putative integration host factor MIHF	NP_215904.1	2
72165	F07	610	Rv2879c	hypothetical protein Rv2879c	NP_217395.1	-
72126	F08	613	Rv3770c	hypothetical protein Rv3770A	YP_178012.1	1.345840131
72299	F09	616	Rv3647c	hypothetical protein Rv3647c	NP_218164.1	2.211038961
72369	F10	643	Rv0612	hypothetical protein Rv0612	NP_215126.1	1.99222395
72417	F11	643	Rv2022c	hypothetical protein Rv2022c	NP_216538.1	2
72663	F12	646	Rv0038	hypothetical protein Rv0038	NP_214552.1	2.578947368
72581	G01	649	Rv0995	ribosomal-protein-alanine acetyltransferase	NP_215510.1	2
72652	G02	652	Rv1210	DNA-3-methyladenine glycosylase I	NP_215726.1	2
72477	G03	652	Rv3055	TetR family transcriptional regulator	NP_217571.1	2
72464	G04	658	Rv0779c	transmembrane protein	NP_215293.1	1.772036474
72549	G05	658	Rv3189	hypothetical protein Rv3189	NP_217705.1	1.993920973
72676	G06	670	Rv2680	hypothetical protein Rv2680	NP_217196.1	1.937313433
72700	G07	673	Rv3153	NADH dehydrogenase subunit I	NP_217669.1	2
72631	G08	679	Rv0175	mce associated membrane protein	NP_214689.1	2.402061856
72415	G09	682	Rv2983	hypothetical protein Rv2983	NP_217499.1	2
72603	G10	682	Rv2986c	DNA-binding protein HU	NP_217502.1	2.315249267
72593	G11	685	Rv0358	hypothetical protein Rv0358	NP_214872.1	1.448175182
72637	G12	685	Rv0956	phosphoribosylglycinamide formyltransferase	NP_215471.1	3.075912409
72501	H01	685	Rv1907c	hypothetical protein Rv1907c		2
72405	H02	685	Rv2506	TetR family transcriptional regulator	NP_217022.1	2
72496	H03	691	Rv0348	transcriptional regulatory protein	NP_214862.1	2
72521	H04	691	Rv2612c	CDP-diacylglycerol-inositol 3-phosphatidyltransferase	YP_177894.1	2
72437	H05	694	Rv0309	hypothetical protein Rv0309	NP_214823.1	2

Clone	Well Position	ORF Length	Locus ID	Description	Accession Number	Average Depth of Coverage
72702	H06	694	Rv1332	transcriptional regulatory protein	NP_215848.1	2
72507	H07	694	Rv1587c	REP13E12 repeat-containing protein	NP_216103.2	3.095100865
72556	H08	694	Rv2086	hypothetical protein Rv2086	NP_216602.2	2
72517	H09	694	Rv2637	transmembrane protein DedA	NP_217153.1	1.998559078
72537	H10	697	Rv2301	cutinase CUT2	NP_216817.2	2
72609	H11	697	Rv2543	lipoprotein LppA	NP_217059.1	2
72498	H12	697	Rv3000	transmembrane protein	NP_217516.1	2.56097561

[1]All information in this table was provided by J. Craig Venter Institute at the time of deposition.